EXHIBIT F

DAY 0. Just before the first challenge.

BAL cellcount 250ul

OX40 -/- = $41 \times 10^4 = 4.1 \times 10^5$ cells/ml.

OX40 -/- = $74 \times 10^4 = 7.4 \times 10^5$ cells/ml.

OX40 +/+ = $75 \times 10^4 = 7.5 \times 10^5$ cells/ml.

OX40 +/+ = $54 \times 10^4 = 5.4 \times 10^5$ cells/ml.

collagenase....

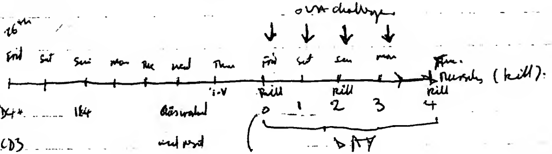
$\frac{5 \text{ mg}}{50} \times 3000 = 300 \mu l$. $\frac{0.1}{10} \times 3000 = 30 \mu l$ DNAse 40 min 37 [illegible]

tissue count :-

FACS

2 mice/group OX40 -/- (2ml) LN = $67 \times 2 \times 10^4 = 1.34 \times 10^6$ cells/ml - (2ml)
(10ml) LUNG = $139 \times 2 \times 10^4 = 2.78 \times 10^6$ cells/ml. (4ml)
(10ml) spleen. = $1036 \times 2 \times 10^4 = 2.072 \times 10^7$ cells/ml. (1ml)

2 mice/group OX40+/+ (2ml) LN = $45 \times 2 \times 10^4 = 9.0 \times 10^5$ cells/ml. (2ml)
(10ml) LUN = $102 \times 2 \times 10^4 = 2.04 \times 10^6$ cells/ml. (4ml)
(10ml) spleen. = $125 \times 2 \times 10^4 = 2080 \times 10^7$ cells/ml. (1ml).



CD4+ ...... IE4   Ößwahed
αCD3   and post
αCD28
αIL-10
αIFN-γ
IL-2
IL4.

mice killed
before the first
challenge.

DAY ②.

BAL cell count => 250 µl.

OX40-/- control = $55 \times 10^4 = 5.5 \times 10^5$

" " #1 = ~~15×10^4~~ $15 \times 10^4 = 1.5 \times 10^5$

#2 = $45 \times 10^4 = 4.5 \times 10^5$

#3 = $8 \times 10^4 = 0.8 \times 10^5$

#4 = $17 \times 10^4 = 1.7 \times 10^5$

OX40+/+ control = $20 \times 10^4 = 2 \times 10^5$

#1 = $114 \times 10^4 = 1.14 \times 10^6$

#2 = $103 \times 10^4 = 1.03 \times 10^6$

#3 = $123 \times 10^4 = 1.23 \times 10^6$

#4 = $115 \times 10^4 = 1.15 \times 10^6$

Dilute,

colony: $\frac{5}{50} \times 5000 = 500\,\mu l$. $\frac{0.1}{10} \times 500 = 50\,\mu l$

Tissue cell count =.

(FACS).

1 mouse { OX40-/- control = (1ml) LN = $210 \times 2 \times 10^4$ — (1ml) 420×10^6 cells/ml

(15ml) LUNG = $37 \times 5 \times 10^4$ — (4ml) 1.85×10^6 cells/ml

(15ml) spleen = $133 \times 5 \times 10^4$ — (1ml) 6.65×10^6 cells/ml X

4 mice (2ml) # 1-4 LN = = ~~83×10^4~~ $148 \times 5 \times 10^4$ — (2ml). 7.40×10^6

3 mice (15ml) # 2-4 LUNG = = $142 \times 5 \times 10^4$ — (0.5ml) 7.10×10^6

2 mice (15ml) # 3-4 spleen = = $273 \times 5 \times 10^4$ — (0.5ml). 1.305×10^7

1 mouse { OX40+/+ control (1ml) LN = $83 \times 2 \times 10^4$ = — (1ml) 1.66×10^6

(15ml) LUNG = $36 \times 5 \times 10^4$ = — (4ml) 1.80×10^6

(15ml) spleen = $141 \times 5 \times 10^4$ = — (1ml). 7.05×10^6

(2ml) # 1-4 LN = $258 \times 5 \times 10^4$ = — (1ml) 1.425×10^7

(15ml) # 2-4 LUNG = $156 \times 5 \times 10^4$ — (1ml) 7.80×10^6

(15ml) # 3-4 spleen = $342 \times 5 \times 10^4$ — (0.5ml). 1.[illegible]

Eosinophil cell count :-

DAY 0	ox40$^{-/-}$ #1	ox40$^{-/-}$ #2	ox40$^{+/+}$ #1	ox40$^{+/+}$ #2.
Total :-	200	200	199	199
Eosinoph :-	0	0	1	1
%	= 0	= 0	= 0	= 0.

DAY 2.	ox40$^{-/-}$ control	ox40$^{+/+}$ control.
Total :-	200	200
Eosinoph :-	0	0.
	= 0	= 0.

ox40$^{-/-}$	#1	#2	#3	#4	ox40$^{+/+}$	#1	#2	#3	#4.
Total	377	very few cells.	209.	265.		463	510	605	468
eosinoph	27	—	28.	66		263	202.	261	303.
% =	7.16%	—	13.40%	24.91%		56.80%	39.61%	43.14%	64.75

BAL cell count 250 ml. DAY 4

OX40-/- control = 64×10^4 = 6.4×10^5.

#1 = $194 \times 2 \times 10^4$ = 3.88×10^6 ~~26~~ 550×250 = 5×2.

#2 = 497×2 = 9.94×10^6.

#3 = 260×2 = 5.20×10^6.

#4 = 240×2 = 4.80×10^6

OX40+/+ control = 65×10^4 = 6.5×10^5.

#1 = $286 \times 2 \times 10^4$ = 5.72×10^6

#2 = $616 \times 2 \times 10^4$ = 1.23×10^7

* #3 = $576 \times 2 \times 10^4$ = 1.15×10^7

#4 = $589 \times 2 \times 10^4$ = 1.18×10^7

tissue count. Collagenase = $\frac{5}{200} \times 6000$ = 300 μl. $\frac{0.1}{10} \times 6000$ = 60 μl.

(1 ml) 1 min OX40-/- = cont. LN = ~~65×10~~ $68 \times 2 \times 10^4$ (1 ml).

(15 ml). 1 min LUNG = $47 \times 2 \times 10^4$ (4.5 ml).

(15 ml). 1 min splen = $246 \times 2 \times 10^4$ (1 ml).

(2 ml) 4 min OVA challenge. #1 - #4 = LN = $700 \times 2 \times 10^4$ = (0.5 ml).

(15 ml) 3 min # ~~1,2,3~~ 2,3,4 = LUNG = $227 \times 5 \times 10^4$ = (1 ml).

(15 ml). 3 min # ~~2~~ 3,4 = spleen = $299 \times 5 \times 10^4$ = (0.5 ml).

~~#~~

(2 ml) ~~1 min~~ 1 min OX40+/+ = control = LN ~~(4,7,3,4)~~ = $103 \times 2 \times 10^4$

(15 ml) 1 min ~~3,3,4~~ 43,4 = LUNG = $49 \times 2 \times 10^4$

(15 ml) ~~3 min~~ 1 min ~~3,3,4~~ splen = $179 \times 2 \times 10^4$.

(2 ml) 3 min OVA challenge. 1,3,4 = LN = $620 \times 2 \times 10^4$ = (0.5 ml).

(15 ml) 3 min 2,3,4 = LUNG = $185 \times 5 \times 10^4$ = (1 ml).

(15 ml). 3 min 43,4 = splen = $337 \times 5 \times 10^4$ = (0.5 ml).

FACS

Folder = BALF - FSCSSC - DAY 4.

BALF DAY 4 FSC/SSC.

	OX	Scan
1.	OX40$^{-/-}$ C	0.01
2.	# 1	0.02
3.	# 2	0.03
4.	# 3	0.04
5.	# 4	0.05
6.	OX40^{++} C	0.06
7.	# 1	0.07
8.	# 2	0.08
9.	# 3	0.09
10.	# 4	0.010
~~11.~~	~~#5~~	
~~12.~~	~~#6~~	

100 10% [illegible]

DAY ④

AG 6756

Investigator: **Croft/Shahram**
Protocol: **AF3-MC1-032802** Rec'd:.
Species: **Mouse** Source: **Jax**
Strain: **C57BL/6J** Sex: **Female**
DOB: Date Weaned:
Dam: Sire:

200ul 2×10^6 cells OT1: Cx40 +/+
ova challenge
ova challenge.
ova challenge

- ova challen
killed

DAY ④

AG 6751

Investigator: **Croft/Shahram**
Protocol: **AF3-MC1-032802** Rec'd:.
Species: **Mouse** Source: **Jax**
Strain: **C57BL/6J** Sex: **Female**
DOB: Date Weaned:
Dam: Sire:

2×10^6 cells OT1: Cx40 -/-
ova challenge.
ova challenge.
ova challenge.

- ova challen
killed

folder 112 :- Adoptin

final settings

100 1. CD4 APC
101 2 Vα PE
102 3. Vβ Cy5. } Before injectn
103 4. CFSE.
104 5. +/+ CFSE
105 6. -/- CFSE.
106 7. control
107 8. CD4 APC.
108 9. Vα
109 10. Vβ

2.29 ⟷ FSC -> 2.05
481 ⟷ SSC $\xrightarrow[\text{FL-1}]{}$ 462.
489 ⟷ FL-1 = 600. ~~474~~ 485
526 ⟷ FL-2 574 520 ~~485~~ 526
718 ⟷ FL-3 - 763 700 718
966 ⟷ FL-4 840 826 .90

FL-1 0.5 FL-2
FL-2 26.5 FL-1
FL-2 0.6 FL-3
FL-3 24.2 FL-2 25.4
FL-3 23.6 FL-4
FL-4 0. FL-3.

111. 110 11. +/+ LN
112 112 111 12. +/+ LUNG
113. 13. +/+ Spleen } DAY 0
114. 14. -/- LN
115. 15. -/- LUNG
116. 16. -/- Spleen

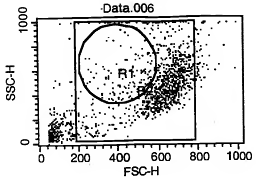
117: 17. +/+ LN
118. 18. +/+ LUNG } Control
119. 19. +/+ Spleen
120. 20. +/+ LN
121. 21. +/+ LUNG } OVA.
122. 22. +/+ Spleen
123. 23. -/- LN
124 24. -/- LUNG } cont
125. 25. -/- Spleen
126. 26. -/- LN
127. 27. -/- LUNG } OVA.
128. 28. -/- Spleen
} DAY 2.

29. +/+ LN
30. +/+ LUNG } cont
31. +/+ Spleen.
32. +/+ LN
33. +/+ LUNG } OVA
34. +/+ Spleen.
35. -/- LN
36. -/- LUNG } cont
37. -/- Spleen
38. -/- LN
39. -/- LUNG } OVA.
40. -/- Spleen
} DAY 4

DAY 2 Total # of VαVβ positive CD4 cells.

		DAY 0	DAY 2.	
~~DAY 0~~	+/+ LN PBS	2.5650×10^{4}	7.40×10^{4}	6.5
	+/+ LN OVA.		4.73×10^{5}	
	-/- LN PBS	2.6667×10^{4}	3.18×10^{5}	
~~DAY 2~~	-/- LN OVA.		2.80×10^{5}	
~~DAY 0~~	+/+ LUNG PBS	2.93760×10^{5}	1.19×10^{6}	
	+/+ LUNG OVA		2.99×10^{6}	7.5
	-/- LUNG PBS	5.27×10^{5}	[redacted] 3.05x10	
	-/- LUNG. OVA.		3.09x10	
~~DAY 0~~	+/+ spleen PBS	184×10^{6}	4.66×10^{6}	
	+/+ spleen OVA		1.40×10^{7}	3-
	-/- spleen PBS	2.76×10^{6}	[redacted] 5.137×10^{6}	
	-/- spleen OVA.		7.32x10	
~~DAY 2~~				

BALF DAY(4).



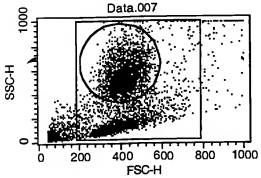
File: Data.006
Acquisition Date:
Gated Events: 4449
X Parameter: FSC-H (Linear)

+/+
Sample ID: 6 (0X40)
Gate: No Gate
Total Events: 4449
Y Parameter: SSC-H (Linear)

Region	Events	% Gated	% Total	X Geo Mean	Y Geo Mean
R1	371	8.34	8.34	443.40	538.14
R2	3433	77.16	77.16	580.35	453.94

0

#1



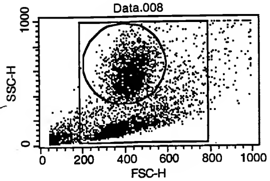
File: Data.007
Acquisition Date:
Gated Events: 20000
X Parameter: FSC-H (Linear)

Sample ID: 7 (0X40+/+)
Gate: No Gate
Total Events: 20000
Y Parameter: SSC-H (Linear)

Region	Events	% Gated	% Total	X Geo Mean	Y Geo Mean
R1	9582	47.91	47.91	416.11	547.33
R2	17592	87.96	87.96	402.27	303.17

54.45

#2



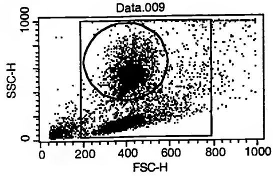
File: Data.008
Acquisition Date:
Gated Events: 20000
X Parameter: FSC-H (Linear)

Sample ID: 8 (0X40+/+)
Gate: No Gate
Total Events: 20000
Y Parameter: SSC-H (Linear)

Region	Events	% Gated	% Total	X Geo Mean	Y Geo Mean
R1	7624	38.12	38.12	420.18	558.87
R2	17364	86.82	86.82	401.27	265.46

44.02

#3



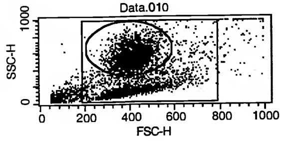
File: Data.009
Acquisition Date:
Gated Events: 20000
X Parameter: FSC-H (Linear)

Sample ID: 9 (0X40+/+)
Gate: No Gate
Total Events: 20000
Y Parameter: SSC-H (Linear)

45

Region	Events	% Gated	% Total	X Geo Mean	Y Geo Mean
R1	7846	39.23	39.23	417.90	547.84
R2	17594	87.97	87.97	403.40	267.84

#4

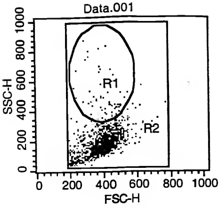


File: Data.010
Acquisition Date:
Gated Events: 20000
X Parameter: FSC-H (Linear)

Sample ID: 10 (0X40+/+)
Gate: No Gate
Total Events: 20000
Y Parameter: SSC-H (Linear)

51.7

Region	Events	% Gated	% Total	X Geo Mean	Y Geo Mean
R1	9063	45.32	45.32	410.59	545.45
R2	17510	87.55	87.55	400.01	298.35

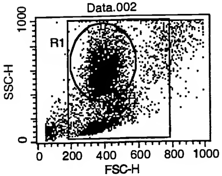


File: Data.001
Acquisition Date:
Gated Events: 1355
X Parameter: FSC-H (Linear)

Sample ID: 1
Gate: G2
Total Events: 2540
Y Parameter: SSC-H (Linear)

Region	Events	% Gated	% Total	X Geo Mean	Y Geo Mean
R1	73	5.39	2.87	399.10	488.51
R2	1355	100.00	53.35	384.48	178.22

#1



File: Data.002
Acquisition Date:
Gated Events: 20000
X Parameter: FSC-H (Linear)

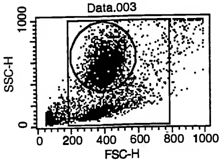
Sample ID: 2 (0X40-/-)
Gate: No Gate
Total Events: 20000
Y Parameter: SSC-H (Linear)

Region	Events	% Gated	% Total	X Geo Mean	Y Geo Mean
R1	9862	49.31	49.31	399.58	570.60
R2	17177	85.89	85.89	405.86	369.43

57.4%

+

#2



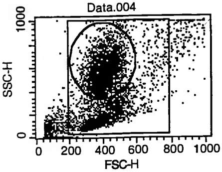
File: Data.003
Acquisition Date:
Gated Events: 20000
X Parameter: FSC-H (Linear)

Sample ID: 3 (0X40-/-)
Gate: No Gate
Total Events: 20000
Y Parameter: SSC-H (Linear)

Region	Events	% Gated	% Total	X Geo Mean	Y Geo Mean
R1	9881	49.41	49.41	409.32	565.83
R2	17406	87.03	87.03	397.05	325.51

56.90

#3



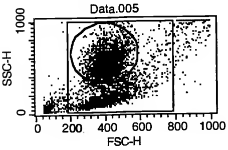
File: Data.004
Patient ID:
Gate: No Gate
Total Events: 20000

Sample ID: 4 (0X40-/-)
Acquisition Date:
Gated Events: 20000

Region	Events	% Gated	% Total	X Geo Mean
R1	10083	50.42	50.42	402.54
R2	17120	85.60	85.60	401.14

58.90

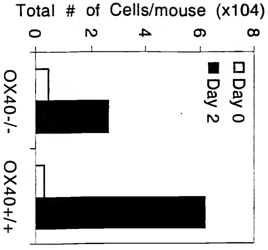
#4

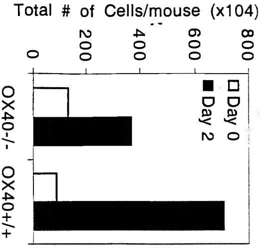


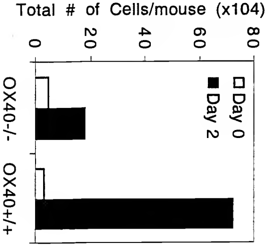
File: Data.005
Acquisition Date:
Gated Events: 20000
X Parameter: FSC-H (Linear)

Sample ID: 5 (0X40-/-)
Gate: No Gate
Total Events: 20000
Y Parameter: SSC-H (Linear)

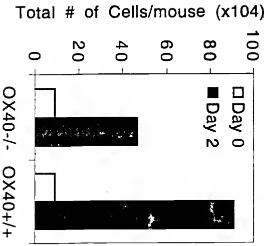
Region	% Gated	% Total	X Geo Mean	Y Geo Mean
R1	49.53	49.53	406.11	536.70
R2	90.44	90.44	403.24	314.07

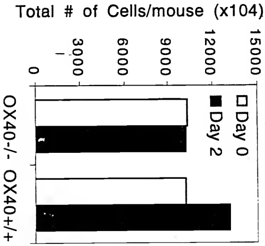

Total # of Cells/mouse (x104)
0
2
4
6
8
Day 0
Day 2
OX40-/-
OX40+/+


Total # of Cells/mouse (x104)
0
200
400
600
800
Day 0
Day 2
OX40-/-
OX40+/+


Total # of Cells/mouse (x104)
0
20
40
60
80
Day 0
Day 2
OX40-/-
OX40+/+


Total # of Cells/mouse (x104)
0
1500
3000
4500
6000
Day 0
Day 2
OX40-/-
OX40+/+

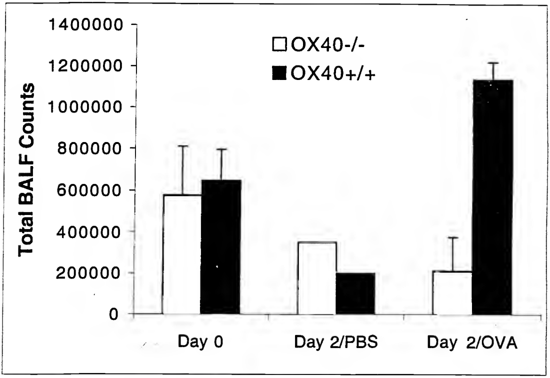

Total # of Cells/mouse (x104)
0
20
40
60
80
100
Day 0
Day 2
OX40-/-
OX40+/+


Total # of Cells/mouse (x104)
0
3000
6000
9000
12000
15000
Day 0
Day 2
OX40-/-
OX40+/+

Total DAY②

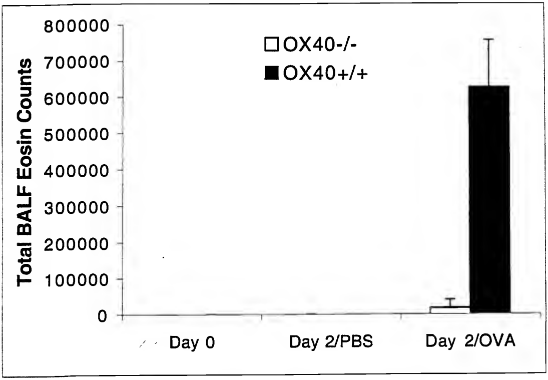
Day0		Day 2			
OX40-/-	OX40+/+	OX40-/-	OX40+/+	OX40-/-	OX40+/+
410000	750000	350000	200000	150000	1140000
740000	540000			450000	1030000
				80000	1230000
				170000	1150000
575000	645000	350000	200000	212500	1137500
233345.238	148492.424	0	0	162967.277	82209.083

	Day 0	Day 2/PBS	Day 2/OVA		
OX40-/-	575000	350000	212500	233345	0
OX40+/+	645000	200000	1137500	148492	0



OX40-/-	Eosinophils%		OX40+/+	Eosinophils%	
#1	#3	#4	#1	#3	#4
150000	80000	170000	1140000	1230000	1150000
7.16	13.4	24.91	56.8	39.61	64.75
10740	10720	42347	647520	487203	744625

	Day 0	Day 2/PBS	Day 2/OVA		
OX40-/-	0	0	18053		0
OX40+/+	0	0	626449	0	0

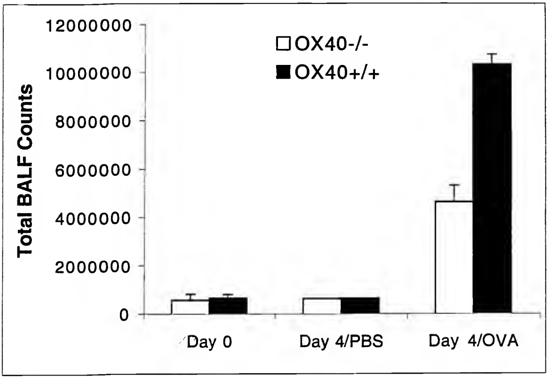


F13

Total DAY④.

Day0		Day 2			
OX40-/-	OX40+/+	OX40-/-	OX40+/+	OX40-/-	OX40+/+
410000	750000	640000	650000	3880000	
740000	540000				12300000
				5200000	11500000
				4800000	11800000
575000	645000	640000	650000	4626666.67	11866666.7
233345.238	148492.424	0	0	676855.474	404145.188

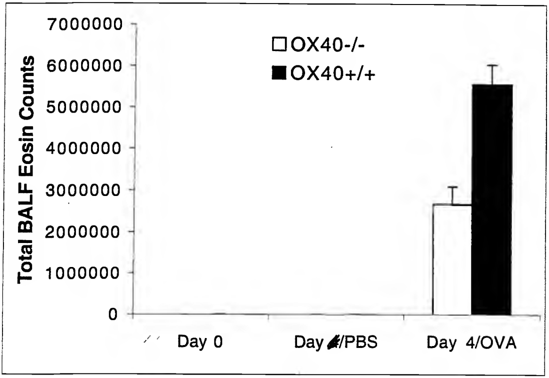
	Day 0	Day 4/PBS	Day 4/OVA			
OX40-/-	575000	640000	4626666		233345	0
OX40+/+	645000	650000	10330000		148492	0



Eosin DAY ④

OX40-/-	Eosinophils%		OX40+/+	Eosinophils%	
#1	#3	#4	#2	#3	#4
3880000	5200000	4800000	12300000	11500000	11800000
57.4	58.9	56.8	44	45	51.76
2227120	3062800	2726400	5412000	5175000	6107680

	Day 0	Day 4/PBS	Day 4/OVA		
OX40-/-	0	0	2672107		0
OX40+/+	0	0	5564893	0	0



OX40-/-	Eosinophils%		OX40+/+	Eosinophils%	
#1	#3	#4		#1	#3	-- #4
150000	80000	170000		1140000	1230000	1150000
7.16	13.4	24.91		56.8	39.61	64.75
10740	10720	42347		647520	487203	744625

Day 2	OX40-/-	OX40+/+	Day 4	OX40-/-	OX40+/+
	7.16	56.8		57.4	54.45
	13.4	39.61		56.8	44
	24.91	64.75		58.9	51.76
	15.1566667	53.72		57.7	50.07
	9.0044452	12.8498911		1.08166538	5.42611279
	OX40-/-	OX40+/+	9	12.85	
Day 2	15.16	53.72	1	5.42	
Day 4	57.7	50.07